



M₃ MUSCARINIC RECEPTOR, HUMAN RECOMBINANT

Expressed in Sf9 Cells

Product Number **M-176**

Product Description

M₃ Muscarinic Receptor, human recombinant (Sf9) is a frozen aliquot of membranes from Sf9 cells transfected with the human recombinant M₃ muscarinic receptor.

Reagent

M₃ Muscarinic Receptor, human recombinant (Sf9) is suspended in 50 mM Tris-HCl, pH 7.4, 10 % glycerol, and 1 % bovine serum albumin (BSA).

Procedure

Incubation buffer:

50 mM Tris-HCl, pH 7.4
10 mM MgCl₂
1 mM EDTA

Binding Protocol

Membranes:

Dilute in incubation buffer (0.5 mL of membrane + 24.5 mL incubation buffer).

Assay mixture:

500 µl diluted membranes
20 µl radioligand
20 µl buffer or unlabeled ligand

Radioligand:

[³H]-N-Methyl scopolamine at a final concentration of 0.1 nM for competition studies.

Unlabeled ligand:

Atropine at a concentration of 1.0 µM.

Incubation time:

60 minutes at 27 °C

Product Information

Separation:

Over GF/C filter (5 mm diam., presoaked in 0.3 % polyethylamine prepared in incubation buffer) then washed 4x with 500 µl of ice cold 50 mM Tris-HCl, pH 7.4.

Results

Typical affinities using standard binding assay above. Results may vary from lot to lot.

Ligand	K _i (nM)
[³ H]-N-Methyl scopolamine	0.19 (Kd)
4-DAMP methiodide	0.87
Pirenzepine	28

Storage/Stability

Store tightly sealed at -80 °C. The membranes will retain their original specific activity for several months when stored at -80 °C in the original packing solution. Repeated freeze-thaw of this product is not recommended.

Precautions

While no human toxicity data is available for this substance, it should be handled with care. Precautions should be taken to avoid contact by all routes of exposure.

References

1. Tobin, A.B. and Nahorski, S.R., *J. Biol. Chem.*, **268**, 9817 (1993).
2. Wall, S.J. et al., *Mol. Pharmacol.*, **40**, 783 (1991).

mje 04/01

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